

First report of *Rhabdochona acuminata* (Nematoda, Rhabdochonidae) in *Astyanax* aff. *fasciatus* (Characiformes, Characidae) from Lake Guaíba, southern Brazil

Primeiro registro de *Rhabdochona acuminata* (Nematoda, Rhabdochonidae) em *Astyanax* aff. *fasciatus* (Characiformes, Characidae) no Lago Guaíba, no sul do Brasil

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Abstract

Specimens of *Rhabdochona* (*Rhabdochona*) *acuminata* (Molin, 1860) were collected in *Astyanax* aff. *fasciatus* (Cuvier, 1819) from Lake Guaíba, municipalities of Guaíba (30°08.28'S, 51°18.53'W) and Barra do Ribeiro (30°17.11'S, 51°18.01'W), southern Brazil. In the present study, the prevalence of *R. (R.) acuminata* was low (1.43%) when compared to other studies performed with *Astyanax* spp. from Brazil. The specimens of *R. (R.) acuminata* showed some morphological differences (mainly the deirids) that have not yet been reported for the species. This is the first report of *R. (R.) acuminata* from Lake Guaíba, filling a gap of occurrence of this nematode in South America.

Resumo

Espécimes de *Rhabdochona* (*Rhabdochona*) *acuminata* (Molin, 1860) foram coletados em *Astyanax* aff. *fasciatus* (Cuvier, 1819) do Lago Guaíba, municípios de Guaíba (30°08.28'S, 51°18.53'W) e, Barra do Ribeiro (30°17.11'S, 51°18.01'W), no sul do Brasil. No presente estudo, a prevalência de *R. (R.) acuminata* foi baixa (1,43%) quando comparada com outros estudos realizados com espécies de *Astyanax* no Brasil. Os espécimes de *R. (R.) acuminata* apresentaram algumas diferenças morfológicas (principalmente nos deirídeos) que não haviam ainda sido registradas para esta espécie. Este é o primeiro registro de *R. (R.) acuminata* para o Lago Guaíba, contribuindo para o conhecimento da distribuição deste nematoide na América do Sul.

Keywords

Characid, morphology, nematode, spirurid, taxonomy

Palavras-chave

Caracídeo, espirurídeo, morfologia, nematoide, taxonomia

Introduction

The subfamily Rhabdochoninae was proposed to group nematodes belonging to the genera *Rhabdochona* Railliet, 1916, *Cystidicola* Fischer, 1798 and *Spinitectus* Fourment, 1883 (Travassos et al. 1928). In the current systematic, this subfamily was elevated to the family level (Gibbons 2010; Moravec 2010).

The genus *Rhabdochona* was proposed to accommodate *Dispharagus denudatus* Dujardin, 1845 (= *Rhabdochona denudata* (Dujardin, 1845) Railliet, 1916), a parasite of *Scardinius erythrophthalmus* (Linnaeus, 1758). Until now, 98 to 104 species were described for this genus, presenting a wide geographical distribution (Mejía-Madrid et al. 2007; Moravec et al. 2013). According to Moravec (2010), the species of *Rhabdochona* originated from a marine ancestral in the Cretaceous period, where regions between seas and rivers alternated incessantly and formed rivers, causing a rapid speciation in host fish and parasites.

In the systematic of the genus *Rhabdochona*, the following subgenera have been proposed: *Filochona* Saidov, 1953, *Rhabdochona* Railliet, 1916 and *Globochona* Moravec, 1972 (Moravec 1972). However, currently most studies agree with the proposal of Moravec (1975), which recognized four subgenera: *Rhabdochona*, *Globochona*, *Globochonoides* Moravec, 1975 and *Sinonema* Moravec, 1975. These subgenera could be differentiated mainly by the number of teeth projection in the prostom, presence or absence of lateral alae, shape of deirids and shape of the tail in the females (Moravec 1975; Moravec 2010).

In the Neotropical Region, 12 species have been reported belonging to the subgenus *Rhabdochona* (Moravec 1998; Ramalho 2005; Moravec 2010), but only *Rhabdochona* (*Rhabdochona*) *acuminata* (Molin, 1860), *Rhabdochona* (*Rhabdochona*) *fabianae* Ramalho, 2005 and *Rhabdochona* (*Rhabdochona*) *uruyeni* Diaz-Ungria,

1968 have been reported from South America. The species of *Rhabdochona* found in South America possibly originated from species found in North America, because all South American species are similar to congeneric species found in Africa (Moravec 2010). The low specificity of the species of *Rhabdochona* from South America could be the reflex of absence of cypriniform fishes, which are considered to be the hosts of several species of *Rhabdochona*, in this geographical region (Mejía-Madrid et al. 2007; Moravec 2010). Besides that, the host distribution explains the morphological differences and the absence of phylogenetic relationships between the South American and African species of *Rhabdochona* (Moravec 2010).

The nematode species *R. (R.) acuminata* was described by Molin in 1860 as *Spiroptera acuminata* in *Brycon falcatus* Müller & Troschel, 1844 from specimens collected by Natterer in 1826 in the state of Mato Grosso, Brazil (Moravec 1972; Cremonte et al. 2002). According to Moravec (2010), *R. (R.) acuminata* is the only species of *Rhabdochona* reported from South America that also could be found in southern Mexico. The species *R. (R.) acuminata* was reported in Ecuador (Petter 1987), in Argentina (Szidat 1956; Cremonte et al. 2002; Ramalho 2005; Ailán Choke et al. 2014) and in Brazil (Travassos et al. 1928; Vaz and Pereira 1934; Kloss 1966; Kohn and Fernandes 1987; Paraguassú et al. 2005; Paraguassú and Luque 2007; Tavernari et al. 2009; Costa et al. 2011; Abdallah et al. 2012; Santos-Clapp and Brasil-Sato 2014; Duarte et al. 2016; Ribeiro et al. 2016; Martins et al. 2017; Yamada et al. 2017; Pereira et al. 2018).

Rhabdochona (R.) acuminata was redescribed by Cremonte et al. (2002) through the examination of specimens collected in fish in Argentina. The present study provides the first record of *R. (R.) acuminata* from southern Brazil and reports some differences found on the morphology hitherto unreported for the species.

Methods

Seventy specimens of *Astyanax* aff. *fasciatus* (Cuvier, 1819) were collected with seine nets between March 2012 and December 2013 from Lake Guaíba, municipalities of Guaíba (30°08.28'S, 51°18.53'W) and Barra do Ribeiro (30°17.11'S, 51°18.01'W), State of Rio Grande do Sul, Brazil. Procedures of transportation and host identification were made according Gallas et al. (2015). Nematodes were collected, fixed, and preserved following Amato and Amato (2010), and they were temporarily mounted and cleared in Amann's lactophenol (Humason 1979).

Measurements are shown in micrometers (µm) unless otherwise stated. The parameters reported are range followed by mean \pm standard deviation, and sample size between parenthesis. Photomicrographs were taken with a Zeiss Axiolab microscope with phase contrast. Line drawings were made with a drawing tube mounted on a Nikon E-200 microscope, scanned and prepared using CorelDraw X4 and Adobe's Photoshop CS2. Ecological parameters follow Bush et al. (1997). Voucher helminth specimens were deposited in the 'Coleção Helmintológica do Instituto Oswaldo Cruz' (CHIOC), Rio de Janeiro, RJ, Brazil.

Results

Rhabdochona (Rhabdochona) acuminata (Molin, 1860)

Figs 1, 2

Description based on 10 specimens. Small to medium nematodes with transverse striated cuticle. Oral aperture oval, with four papillae, two ventral and two dorsal. Vestibule elongated, dilated anteriorly to form a funnel-shaped prostom. Prostom anteriorly armed with 14 longitudinal projections similar to teeth in the inner surface. Prostom with basal tooth. Simple deirids, anterior to the nerve ring and the excretory pore. Esophagus divided in muscular and glandular portions, of which the muscular is the smaller portion. Tail of both sexes conical, sharply pointed.

Males ($n = 3$). Body 16.5–18.2 mm (17.6 ± 0.9 mm; $n = 3$) long, 0.1–0.2 mm (0.14 ± 0.06 mm; $n = 3$) wide. Prostom 40–42 (41 ± 1 ; $n = 3$) long, 27–30 (29 ± 1 ; $n = 3$) wide. Vestibule (including prostom) 127–137 (134 ± 6 ; $n = 3$) long, 12–15 (14 ± 1 ; $n = 3$) wide. Deirids 40–45 (42 ± 2 ; $n = 3$) from anterior extremity. Nerve ring and excretory pore 192–232 (206 ± 23 ; $n = 3$) and, 330–350 (339 ± 10 ; $n = 3$) from anterior extremity, respectively. Muscular esophagus 480–600 (500 ± 92 ; $n = 3$) long, 50 wide; glandular esophagus 4.7–5 mm (4.9 ± 0.1 mm; $n = 3$) long, 0.12–0.17 mm (0.15 ± 0.03 mm; $n = 3$) wide. Esophagus length representing 28.12% of total body length (TBL); muscular esophagus corresponding to 10.1% of glandular esophagus. Posterior extremity with 16 pairs of papillae: 10 pairs precloacal, of which the fourth pair (from cloaca to the anterior extremity) is more lateral; six

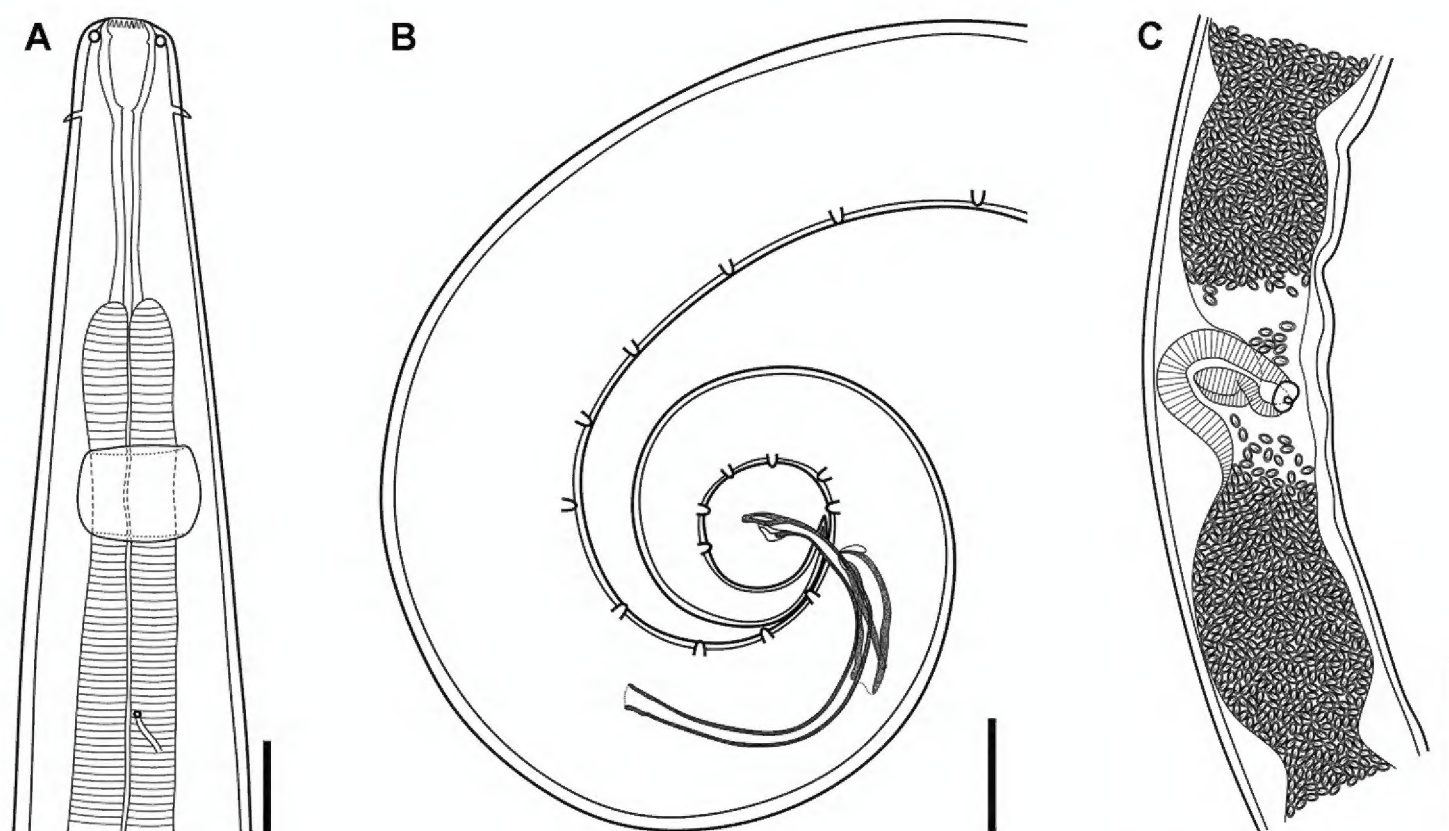


Figure 1. Incomplete diagrams of *Rhabdochona (Rhabdochona) acuminata* (Molin, 1860) from Lake Guaíba, southern Brazil. **A.** Male anterior portion, ventral view. **B.** Male posterior end, lateral view. **C.** Region of the vulva in the female, ventral view. Scale bars: 50 μ m (**A**), 100 μ m (**B**), 350 μ m (**C**).

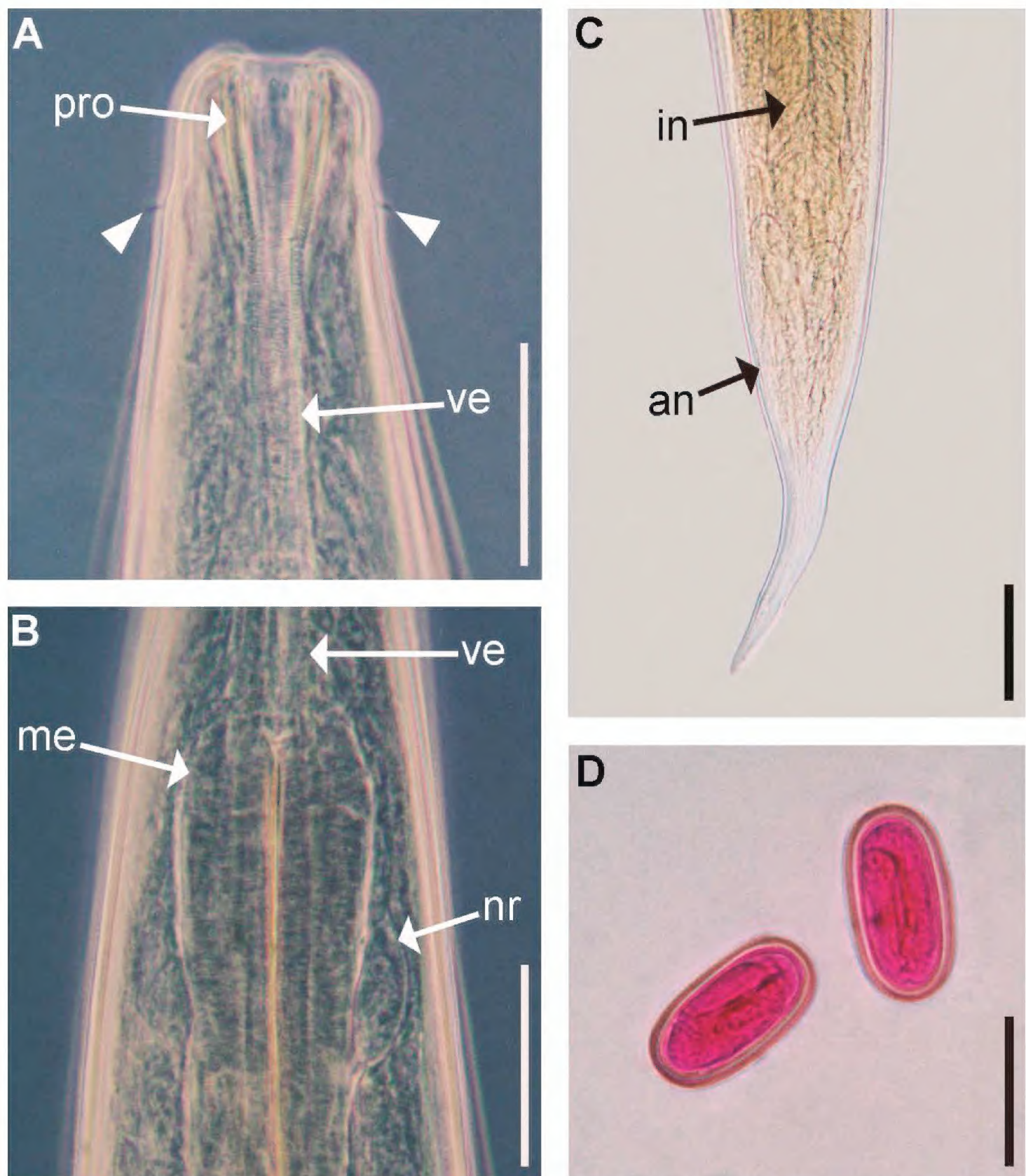


Figure 2. Photomicrographs of *Rhabdochona* (*Rhabdochona*) *acuminata* (Molin, 1860) from Lake Guaíba, southern Brazil. **A.** Female anterior end seen with phase contrast, ventral view, showing prostom (pro), vestibule (ve) and deirids (white arrowheads). **B.** Female anterior end seen with phase contrast, ventral view, showing vestibule (ve), muscular esophagus (me) and nerve ring (nr). **C.** Female posterior region, lateral view, showing intestine (in) and anus (an). **D.** Eggs stained with rose bengal. Scale bars: 50 µm (A, B), 100 µm (C), 25 µm (D).

pairs poscloacal, of which the second pair (from cloaca to the posterior extremity) is more lateral. Unequal spicules, the right smaller, 130–142 (136 ± 6 ; $n = 3$) long, and the left 395–445 (420 ± 25 ; $n = 3$) long. Cloaca 320–440 (391 ± 25 ; $n = 3$) from posterior extremity.

Females (n = 7). Body 35.3–41.8 mm (38 ± 2 mm; n = 7) long, 0.3–0.4 mm (0.4 ± 0.03 mm; n = 7) wide. Prostom 45–52 (49 ± 2 ; n = 7) long, 22–37 (30 ± 6 ; n = 7) wide. Vestibule (including prostom) 135–160 (145 ± 9 ; n = 7) long, 12–20 (15 ± 3 ; n = 7) wide. Deirids 32–42 (38 ± 3 ; n = 7) from anterior extremity. Nerve ring and excretory pore 170–270 (206 ± 32 ; n = 7) and 272–312 (299 ± 16 ; n = 5) from anterior extremity, respectively. Muscular esophagus 550–610 (576 ± 26 ; n = 7) long, 50–60 (54 ± 5 ; n = 7) wide; glandular esophagus 2.8–5.9 mm (5.2 ± 1 mm; n = 7) long, 0.1–0.2 mm (0.18 ± 0.04 mm; n = 7) wide. Esophagus length representing 15.4% of TBL; muscular esophagus corresponding 10.9% of glandular esophagus. Amphidelphic uterus and post equatorial vulva, 18.4–22.4 mm (20.8 ± 1.7 mm; n = 5) from anterior extremity. Eggs smooth, nonfilamented and embryonated, 32–37 (36 ± 1 ; n = 30) long, and 17–22 (19 ± 1 ; n = 30) wide. Anus 340–420 (370 ± 30 ; n = 7) from posterior extremity.

Taxonomic summary:

Synonyms: *Spiroptera acuminata* Molin, 1860, *Rhabdochona elegans* Travassos, Artigas & Pereira, 1928, *Rhabdochona fasciata* Kloss, 1966, *Rhabdochona australis* Kloss, 1966, *Rhabdochona siluriformis* Kloss, 1966.

Host: *Astyanax* aff. *fasciatus* (Cuvier, 1819).

Locality: Municipality of Guaíba, Lake Guaíba (30°08.28'S, 51°18.53'W), RS, Brazil.

Site of infection: Anterior intestine.

Prevalence: 1.43%.

Mean intensity of infection: 13 helminths/host.

Mean abundance of infection: 0.19 helminth/host.

Voucher specimens of helminths deposited: CHIOC 35947.

Discussion

The measures of *R. (R.) acuminata* found in the literature until 2002 were compiled and compared by Cremonte et al. (2002). Posteriorly, measurements of *R. (R.) acuminata* were taken by Ramalho (2005) in Argentina and, Costa et al. (2011) in Brazil. The specimens examined in the present study showed similar measurements when compared with the tables organized by Cremonte et al. (2002). However, males and females of *R. (R.) acuminata* found in *A. aff. fasciatus* from Lake Guaíba presented larger specimens when compared to the specimens reported by Cremonte et al. (2002), Ramalho (2005) and Costa et al. (2011) (Tables 1, 2). Besides that, the table organized by Cremonte et al. (2002) possibly has a scale error, because the width measurements of males and females (62–300 mm) and other traits are higher than the variation in the measurements of length (4.54–28 mm).

According to Cremonte et al. (2002), the longest length recorded to *R. (R.) acuminata* was 13 mm for males (Table 1) and 28 mm for females (Table 2). In the present study, males were 16.5 to 18.2 mm (17.6 mm) long and females 35 to 42 mm (38 mm) long. Moreover, the deirids observed in the specimens collected

Table 1. Comparison of measurements (mm) of male specimens of *Rhabdochona* (*Rhabdochona*) *acuminata* (Molin, 1860) obtained from different studies.

Reference	Travassos et al. (1928)	Vaz and Pereira (1934)	Kloss (1966)	Moravec (1972)	Cremonte et al. (2002)	Ramalho (2005)	Costa et al. (2011)	Present study
Body length	8–13	8.9	4.54–8.78	6.3–10.22	4.18–9.19	4.94–8.24	6.1–8.8	16.5–18.2
Body width	0.1–0.2	0.12	0.06–0.12	0.08–0.12	0.07–0.13	0.08–0.11	0.09–0.13	0.1–0.2
Prostom	–	0.06 × 0.04§	–	0.01–0.03 × 0.009–0.015	0.01–0.03 × 0.01–0.02	0.01–0.02 × 0.01	0.02 × 0.01	0.04 × 0.03
Vestibule length	0.1–0.15§	0.18	0.11–0.14	0.12–0.15	0.08–0.16	0.1–0.14	0.12–0.13	0.13–0.14
Deirids*	–	–	–	0.01–0.05	0.03–0.06	0.04–0.07	0.02–0.03	0.04
Nerve ring*	–	0.24§	0.1–0.25	0.15–0.21	0.13–0.2	0.13–0.21	0.15–0.17	0.2–0.23
Excretory pore*	0.25§	0.41§	0.26	0.24	0.17–0.31	0.18–0.28	0.18–0.19	0.33–0.35
Muscular esophagus	0.44–0.47§	0.32	0.23–0.33	0.27–0.36	0.2–0.37	0.2–0.31	0.29–0.35	0.48–0.6
Glandular esophagus	4.8–5.2§	1.04	1.1–1.63	1.18–1.96	0.85–1.83	0.75–1.54	1.9	4.7–5
Right spicule	0.13–0.14	0.14	0.07–0.13	0.08–0.12	0.07–0.1	0.07–0.08	0.11–0.12	0.13–0.14
Left spicule	0.42–0.44	0.34	0.35–0.49	0.56–0.59	0.42–0.49	0.38–0.43	0.37–0.39	0.39–0.44
Tail length**	0.35–0.40	0.2	0.21–0.37	0.24–0.37	0.25–0.38	0.2–0.28	0.29–0.3	0.32–0.44

§ not specified if male or female;
* distance from anterior extremity;
** distance from posterior extremity.

Table 2. Comparison of measurements (mm) of female specimens of *Rhabdochona* (*Rhabdochona*) *acuminata* (Molin, 1860) obtained from different studies.

Reference	Travassos et al. (1928)	Vaz and Pereira (1934)	Kloss (1966)	Moravec (1972)	Cremonte et al. (2002)	Ramalho (2005)	Costa et al. (2011)	Present study
Body length	22–28	18	12.61–22.3	10.7–20.4	7.45–16.55	5.11–15.2	11–14.6	35.3–41.8
Body width	0.3	0.28	0.12–0.29	0.09–0.27	0.1–0.2	0.1–0.2	0.17–0.22	0.3–0.4
Prostom	–	0.06 × 0.04§	–	0.02–0.03 × 0.01–0.03	0.02–0.03 × 0.01–0.02	0.02–0.03 × 0.01–0.02	0.02–0.03 × 0.02	0.04–0.05 × 0.02–0.04
Vestibule length	0.1–0.15§	0.13	0.12–0.16	0.14–0.15	0.1–0.19	0.11–0.15	0.11–0.13	0.13–0.16
Deirids*	–	–	–	0.06	0.04–0.07	0.05–0.12	0.03–0.04	0.03–0.04
Nerve ring*	–	0.24§	0.21–0.37	0.2–0.27	0.13–0.23	0.14–0.2	0.13–0.22	0.17–0.27
Excretory pore*	0.25§	0.41§	0.28–0.41	–	0.17–0.4	0.15–0.28	0.21–0.27	0.27–0.31
Muscular esophagus	0.44–0.47§	0.6	0.39–0.54	0.32–0.47	0.22–0.44	0.25–0.4	0.32–0.45	0.55–0.61
Glandular esophagus	4.8–5.2§	2.6	1.72–5.14	1.44–4.41	1.05–1.91	1.23–1.64	2.3–3.8	2.8–5.9
Vulva**	–	9.25	5.06–11.1	4.27–8.19	3.35–7.7	3.8–7.36	5.2–8	18.4–22.4
Eggs	0.04 × 0.02	–	0.01 × 0.02	0.03 × 0.02	0.02–0.03 × 0.01–0.02	0.02–0.03 × 0.01–0.02	0.03 × 0.02	0.03–0.04 × 0.02
Tail length**	0.38	0.36	0.21–0.29	0.22–0.24	0.17–0.25	0.19–0.26	0.23–0.24	0.34–0.42

§ not specified if male or female;
* distance from anterior extremity;
** distance from posterior extremity.

here were in a higher position (next to the basal tooth in the prostom), in relation to other studies where the deirids were reported below to the basal tooth (Moravec 1972; Cremonte et al. 2002; Ramalho 2005). These differences could be related to the larger specimens examined in the present study or, to a morphological variation hitherto unreported.

The morphology of the distal region of the left spicule in the species of *Rhabdochona* is complex and may present differences according to the position in which it is examined (Moravec 2010). In the present study, the morphology of the right spicule (with membranous ala on distal end) and the left spicule (with membranous heel and dorsal hook with folded tip) were similar to those described by Travassos et al. (1928), Vaz and Pereira (1934), and more detailed by Cremonte et al. (2002)

and Ramalho (2005). In the species grouped in the subgenus *Rhabdochona* the right spicule possibly is associated with the function of a gubernaculum, which is absent in these species (Moravec 2010).

The prevalence of *R. (R.) acuminata* observed in *A. aff. fasciatus* (n = 70) collected between March 2012 and December 2013 here was extremely low (1.43%) when compared to the prevalence reported in *Astyanax bimaculatus* (Linnaeus 1758) (41%) (n = 39, between April 2002 and July 2003) and *A. fasciatus* (36.7%) (n = 79, between April 2002 and July 2003) from the state of Rio de Janeiro (Paraguassú and Luque 2007). Such differences could be associated with characteristics in the life cycle of *R. (R.) acuminata* and in the environments. The life cycle of the species of *Rhabdochona* found in South America is still unknown, but may present similarities with the life cycles of the other species of the genus, where aquatic insects (ephemeropterans, trichopterans and plecopterans) are considered to be the intermediate hosts (Moravec 2010). The low prevalence found in *A. aff. fasciatus* here could be associated with the feeding habit and the diet of this characid in Lake Guaíba, where the availability and/ or the ingestion of the insects considered intermediate hosts is smaller than the characids examined in the Rio de Janeiro State (Paraguassú and Luque 2007). Besides that, the period of host collection, pollution and the water regime may influence the prevalence of *R. (R.) acuminata*.

In Brazil, there are reports of *R. (R.) acuminata* in the following species of *Astyanax*: *A. bimaculatus*, *A. fasciatus* and *Astyanax schubarti* Britski, 1964 from the state of São Paulo (Kloss 1966) and *A. bimaculatus* and *A. fasciatus* from the state of Rio de Janeiro (Paraguassú and Luque 2007). This is the first report of *R. (R.) acuminata* in *A. aff. fasciatus* from Lake Guaíba, southern Brazil. This study extends the known geographical distribution of *R. (R.) acuminata* and contributes to the knowledge of the helminth fauna of *A. aff. fasciatus*.

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